Pharmaceuticals and Personal Care Products in Biosolids/Sewage Sludge - The Interface between Analytical Chemistry and Regulation

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[Note: minor content and formatting differences may exist between this web version and the published version.]

Web-version of article published: Anal Bioanal Chem; DOI 10.1007/s00216-006-0942-z
Received: 31 July 2006 / Revised: 2 October 2006 / Accepted: 12 October 2006; Printed on-line: 28-Nov-2006

Abstract
Modern sanitary practices result in large volumes of human waste, as well as domestic and industrial sewage, being collected and treated at common collection points, wastewater treatment plants (WWTPs). In recognition of the growing use of sewage sludge as fertilizers and as soil amendments, and the scarcity of current data regarding the chemical constituents in sewage sludge, the United States National Research Council (NRC) in 2002 produced a report on sewage sludge. Among the NRC's recommendations was the need for investigating the occurrence of pharmaceuticals and personal care products (PPCPs) in sewage sludge. PPCPs are a diverse array of non-regulated contaminants that had not been studied in previous sewage sludge surveys but which are likely to be present. The focus of this paper will be to review the current analytical methodologies available for investigating whether pharmaceuticals are present in WWTP-produced sewage sludge, to summarize current regulatory practices regarding sewage sludge, and to report on the presence of pharmaceuticals in sewage sludge.

Web-version of article published: Anal Bioanal Chem; DOI 10.1007/s00216-006-0942-z
Received: 31 July 2006 / Revised: 2 October 2006 / Accepted: 12 October 2006; Printed on-line: 28-Nov-2006

Introduction

Modern sanitary practices result in large volumes of human waste, as well as domestic and industrial sewage, being collected and treated at common collection points, wastewater treatment plants (WWTPs). WWTPs produce aqueous effluents for discharge back into the environment, and sewage sludge. Sewage sludge is usually further treated before use as fertilizers and soil amendments, or disposed as waste. Sewage sludge can be defined as the solid semi-solid residue left over after the treatment of wastewater. It needs to be stated up front that in the United States the term biosolids is used interchangeably with the term sewage sludge, although it is the latter term that is defined in United States (U.S.) statute. Sewage sludge in the U.S. is classified as either Class A or Class B [1]. Class A sewage sludge is defined as sewage sludge that have undergone treatment to reduce pathogens, including pathogenic bacteria, enteric viruses, and viable helminthes ova, below detectable levels as set by EPA's 40 CFR Part 503 [1]. Class A sewage sludge (but not Class B) can be distributed for use as a soil amendment without imposing site and harvesting restrictions. Some examples of the treatment processes used to meet the Part 503 Biosolids Rule Class A pathogen reduction requirements include composting, heat drying, and high-temperature aerobic digestion [1]. Class B sewage sludge is defined as sewage sludge in which the pathogens have been reduced in density. Class B sewage sludge may still contain some pathogens, therefore there are site restrictions required that restrict crop harvesting, animal grazing, and public access for a period of time after application. Unlike Class A sewage sludge, Class B sewage sludge cannot be sold or given away in a bag or other container for land application at public contact sites (e.g., parks, golf courses, lawns, and home gardens). Class B sewage sludge can be used in bulk at appropriate types of land application sites, such as agricultural lands, forests, and reclamation sites, if the sewage sludge meets the limits on metals, vector attraction reduction, and other management requirements of 40 CFR Part 503 [1].

In 1998, approximately 6.9 million tons of sewage sludge was generated in the U.S., of which 60 percent was land-applied (e.g., used as landfill cover, used as fertilizer for silviculture and pasture, or used as a soil amendment in land reclamation) and 40 percent disposed of (i.e., 22 percent incinerated and the other 18 percent discarded *in toto* to landfills) [2]. It is further estimated that at least 20 percent of sewage sludge was managed by municipal solid waste (MSW) facilities through either landfilling (17 percent) or as landfill cover (3 percent). It was expected in 1998 that the volume of sewage sludge being land-applied would increase to 8.2 million tons by 2010 [2]. However, according to the latest data available in 2000 only 45% of sewage sludge was made use of through land-application (40%) or composting (5%), while 45% were disposed of via landfills (17% of the total), incineration (22%), or "other" methods (7%), and the remaining 8% were disposed of via surface disposal or lagoon storage [3].

In recognition of the growing use of sewage sludge and the scarcity of current data regarding the chemical constituents in biosolids, the United States National Research Council (NRC) produced a report on biosolids in 2002 [4]. Among the NRC's recommendations was the need to investigate the occurrence of pharmaceuticals and personal care products (PPCPs) in biosolids. PPCPs are a diverse array of non-regulated chemicals that had not been assessed in previous biosolids surveys, but which are likely to be present.

There are very few studies in the literature regarding the analysis of pharmaceuticals in raw and processed sewage sludge. The focus of this paper will be: (1) a review of the current analytical methodologies available for the analysis of pharmaceuticals in biosolids/sewage

sludge; (2) current regulatory practices in the U.S. and Europe regarding biosolids/sewage sludge; and (3) report on the presence of pharmaceuticals that have been detected in biosolids/sewage sludge.

Sewage sludge production

Wastewater influent from domestic and some industrial sources undergo preliminary, primary, secondary, and in some cases tertiary treatment before sewage sludge is produced and the final effluent is discharged. Initially, the influent is screened to remove large (> 1/2") materials (e.g., rocks, pieces of glass and plastic, sticks). Solids are settled out in primary and/or secondary settling tanks. The solids then undergo further treatment: 1) thickening 2) stabilization 3) conditioning and 4) de-watering. The solid materials are flocculated from the water (thickening) via gravity or dissolved air flotation. The next step, stabilization, generally occurs through the use of anaerobic and aerobic digesters. Anaerobic digestion reduces the volatile solid content by approx. 60 to 65%, and significantly reduces pathogens. The digested sewage sludge, at about 2% solids, is then chemically conditioned by the addition of inorganic or organic chemicals (inorganic: ferric chloride or lime; organic: polymers), and finally, de-watered, to produce a final sewage sludge product (see Figure 1) [5,6]. A significant number of WWTPs send de-watered sewage sludge to compost operations, where the sludge is composted under aerobic conditions with greenwaste or other bulking agents to achieve a compost of about 50% solids, or to heat drying facilities, which dry them to 95% solids for use as fertilizer or fuel. The quantity and characteristics of the sewage sludge generated at a WWTP depends upon the composition of the wastewater, the type of wastewater treatments (i.e., primary, secondary, tertiary), and the types of subsequent treatments applied to the sewage sludge. Even within an individual plant, the characteristics of the sewage sludge produced can change annually, seasonally, or daily because of variations in the composition of the incoming wastewater and variations in the day-to-day treatment processes [2]. This diverse and changing makeup of sewage sludge presents an added challenge for chemical characterization by analytical chemistry.

Analytical Challenges

Sewage sludge can comprise WWTP-produced material from human waste (a mix of excreta containing bacterial microflora, fats, proteins, pigments, and ingested xenobiotics such as pharmaceuticals and illicit drugs, excreted unchanged or as metabolites), along with other domestic and industrial wastes, or it can be a mix of WWTP-produced sewage sludge, along with organic municipal solid waste (e.g., yard trimmings or other greenwastes). The sewage sludge produced for land application can be in a dry pelletized form (95% solids), composted (~ 50% solids), in cake form (~ 15-30% solids), or a semi-liquid form (7-10% solids), depending upon how the sewage sludge is to be distributed and used (e.g., consumer use, or large-scale agricultural use, including farms, orchards, public golf courses, re-forestation), method of incorporation (e.g., sprayed, injected below surface), and the distances it is to be transported prior to use or disposal.

All forms of sewage sludge have physical properties that pose challenges for analytical chemistry methods development, including particles with large surface areas (0.8 - 1.7 m^2/g), negative surface charges, and interstitial spaces, all of which promote sorption, foster occlusion into the biomass, and strong bonding between charged species and the surfaces. Additional challenges are created by the chemical additives used in conditioning step, including ferric chloride, lime, and cationic polyacrylamide polymers (the most widely used polymer for conditioning) [5]. Rogers [7] reported that during primary sedimentation, hydrophobic

chemicals may partition to settled primary sludge solids, and this can be correlated with a chemical's octanol-water partition coefficient (K_{ow}). The following can be used as a general guide: log K_{ow} < 2.5 yields low sorption potential, log K_{ow} > 2.5 and < 4.0 yields medium sorption potential, and log K_{ow} > 4.0 promotes high sorption potential. In 2002, Khan and Ongerth [8] published a fugacity-based model as a useful tool for predicting concentrations of pharmaceutical residuals in primary and secondary sewage sludge.

PPCPs present in biosolids/sewage sludge

In the 1980's and 90's, the focus on the analysis of sewage sludge mainly dealt with pathogens, inorganics (metals), polynuclear aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), and polychlorinated dibenzo-p-dioxins and dibenzofurans (PCDD/Fs) [9-12]. However, recent advances in environmental analytical sciences have revealed a wide variety of pharmaceuticals in wastewaters and surface waters, leading scientists to hypothesize that pharmaceuticals may also be present in sewage sludge [13-18].

There are few reports of pharmaceuticals detected in sewage sludge. Rogers [7] presents one of the first reports to assert the potential for pharmaceuticals to occur in sewage sludge, by assessing the biodegradability of selected pharmaceuticals and pointing to an earlier finding by Richardson and Bowron [19] of aspirin, caffeine, methaqualone and methotrexate detected in sewage, sewage effluent, and source waters.

In 2003, researchers in the United Kingdom (UK) conducted a survey of sewage sludge from 14 WWTPs, not only for select traditional analytes [i.e., PAHs, PCBs, polychlorinated naphthalenes (PCNs - structurally similar to PCBs, several of which exhibit dioxin-like toxicity), polychlorinated n-alkanes (PCAs)], and for a class of PPCPs - synthetic musks [20]. In their survey, the most abundant synthetic musks found were Galaxolide (HHCB) and Tonalide (AHTN), at levels of 27 mg/kg and 4.7 mg/kg, respectively. These values are comparable to those found by Osemwengie [21] (18 mg/kg HHCB and 4.0 mg/kg AHTN, highest concentration found), and Kupper et al. [22] (20 mg/kg HHCB and 7 mg/kg AHTN). Other authors have also reported nonylphenols (NPs), alkylphenols (APs), and synthetic musks, in sewage sludge from both Europe and the US [23-27].

Kim et al. [28] conducted a study on the removal of the antibiotic, tetracycline, during wastewater treatment, in the activated sludge process. The occurrence of tetracycline's were imputed in the sewage sludge by calculating the adsorption kinetics, K_{ads}, of about 8400 ± 500 mL/g. With this value it was predicted that a significant portion of the tetracyclines in wastewater sorbs onto the sewage sludge, and consequently may become bioavailable when the sewage sludge is land-applied [28].

Jacobsen and Halling-Sørensen [29] developed a pressurized liquid extraction (PLE) method (three-step extraction using two different extraction buffers; no pre-heat; 5 min heat; extraction temperature 75^{o}C and pressure 2500psi; 50% flush volume; 60-sec purge) followed by detection with liquid chromatography-electrospray ionization-mass spectrometry-mass spectrometry (LC-ESI-MS/MS) for three classes of antibiotics - tetracyclines, sulfonamides, and tylosin (a macrolide) - in swine manure. They report concentrations of tetracyclines up to 30 mg/kg (dry weight) and sulfonamides up to 2 mg/kg (dry weight), but tylosin was not detected in any manure samples [29]. The findings of large amounts (ppm) of tetracyclines in manure support the predictions of Kim et al. [28] regarding the sorption kinetics of tetracycline.

Gölet, et al. [30] report fluoroquinolones in the concentration range of 1 to 4 mg/kg in sewage sludge from Switzerland. Sewage sludge was extracted using PLE (aqueous 50 mM phosphoric acid with acetonitrile [1:1]; extraction temperature 100°C and pressure 1450 psi; preheat 5min; static 15min; final volume 22 mL; 150% flush volume, cycles: 4 or 6) followed by solid phase extraction (SPE) and analysis by liquid chromatography-fluorescence detection (LC-FLD).

Khan and Ongerth [8] report on the concentrations of six pharmaceuticals in both primary and secondary sludge: paracetamol, naproxen, salicylic acid (ex-aspirin), gemfibrozil, ibuprofen, and carbamazepine. The concentrations reported range from not detected (gemfibrozil) to 0.01 µg/kg (carbamazepine), dry weight. The analytical methodology included soxhlet extraction, followed by evaporation, derivatization and analysis by GC-MS, but no details or experimental recovery data are provided.

Recently, the topical antiseptic agent, triclocarban, has been reported in sewage sludge. The amounts calculated in the sewage sludge exceed more than one metric ton (1000 kg) per year, attributable to just one WWTP. Sewage sludge was extracted using PLE, with the following conditions: extracting solvent acetone; extraction temperature 100°C; pressure1500 psi; static 5min; 60% flush volume, cycles: 1. Analysis of the extracts was with negative ionization LC-ESI-MS/MS [31].

Göbel et al. [32] report on the extraction and determination of various antibiotics including sulfonamides (sulfapyridine, sulfamethoxazole), macrolides (azithromycin, roxithromycin, clarithromycin), and trimethoprim, in dried samples of activated and digested sewage sludge. Two different extraction methodologies were investigated, ultrasonic solvent extraction (USE) and PLE. PLE was chosen over USE as the better extraction methodology. PLE conditions were: extracting solvents methanol:water (1:1); extraction temperature 100°C; pressure 1450 psi; static 5 min; preheat 5 min; 120% flush volume, cycles: 3. Post-extraction clean-up of the extracts with SPE was followed with analysis by LC-ESI-MS/MS. Recoveries of analytes from activated and digested sewage sludge ranged (dry wt) from not detected (nd) to 197 µg/kg sulfapyridine, 113 µg/kg sulfomethoxazole, 133 µg/kg trimethoprim, 158 µg/kg azithromycin, 63 µg/kg clarithromycin, and 131 µg/kg roxithromycin [32].

Kinney et al. [33] have reported on the analysis of nine different sewage sludge products (both Class A and Class B biosolids) for 16 pharmaceuticals, as well as for a variety of non-pharmaceutical organic compounds. For the polar pharmaceuticals, the sewage sludge was extracted using PLE. The following conditions were used: extracting solvent acetonitrile/water mixture (70:30, v/v); extraction temperature 130°C; pressure 1490 psi; static 10 min; cycles: 5. Analyses were performed using external standard calibration by LC-ESI-MS. Three pharmaceuticals were detected in all nine sewage sludge samples: carbamazepine, fluoxetine, and diphenhydramine [33]. The other thirteen pharmaceuticals (acetaminophen, albuterol, dehydronifedipine, diltiazem, gemfibrozil, sulfamethoxazole, thiabendazole, trimethoprim, warfarin, cimetidine, codeine, miconazole) were detected in varying amounts, ranging from not detected to ppb levels, in the nine sewage sludge samples [33].

Research efforts at the U.S. Environmental Protection Agency (US EPA) National Exposure Research Laboratory in Las Vegas, Nevada, have provided for EPA's Office of Water

extraction and detection methods for three classes of PPCPs in two Class A biosolids (Milorganite® and Southern California Los Angeles Hyperion WWTP). The methods include two antibiotic classes - fluoroquinolones (ciprofloxacin and norfloxacin) and macrolides (azithromycin, roxithromycin, and clarithromycin), and synthetic musks [21]. The synthetic musk technique has already been published [21]. We will report here on the development and results from the macrolide antibiotic methodology. Both USE and PLE were investigated as extraction techniques. Three materials, sand, Milorganite® (commercially available Class A biosolids from Milwaukee, WI) and a Class A biosolids from the Southern California Los Angeles Hyperion WWTP were tested. The recoveries were very different depending on the material tested. Lower recoveries were obtained from USE than from PLE, therefore only the PLE methodology will be discussed. Because of the complexity and variable sizes of biosolids particulates, they need to be homogenized before extraction. Batches of biosolids can be pre-dried if in semi-liquid form before homogenization can proceed. Either a 0.5 g or 1 g sample of biosolids is placed in individual smooth-surface porcelain mortars. The dried biosolids are then ground using a pestle. A resulting fine powder material is then ready for extraction. The PLE methodology used an Accelerated Solvent Extraction (ASE) system (Model ASE200, Dionex Corporation, Sunnyvale CA); the conditions were as follows: 99% methanol/1% acetic acid as the extracting solvent; 2-cycles; 2800 psi; extraction temperature: 50°C. The extracts are evaporated under nitrogen (23°C, 5 psi, Zymark TurboVap) to 5 mLs, at which point the extracts are removed from the TurboVap and washed with hexane three times (this step removes most of the extracted lipid material). The extract is then placed back in the TurboVap and evaporated to 0.5 mL. The resultant extract is analyzed by LC-ESI-MS/MS. Data were acquired in the positive ionization mode with a Classic LCQ ion trap mass spectrometer (ThermoFinnigan, San Jose, CA). Because of the extremely large amounts of interfering materials co-extracted with the pharmaceuticals, the analyses were performed using the MS/MS mode for both identification and quantitation of the three macrolides. Table 1 shows the product ions and collision energies used to identify and quantify the macrolides. HPLC separations were performed using an Agilent Zorbax RX-C_{18}, 3.5 µm particle size, 100 x 2.1 mm liquid chromatography column (Agilent Technologies, Santa Clara, CA), with a flow rate of 0.10 mL/min, with a 40:60 split after the column, such that 40% of the flow (40 µL/min) goes to the ES-ITMS. For example, if the injection volume on-column was 10 µL, then the volume entering the ES-ITMS is only 4 µL, due to the 40:60 split. The gradient elution conditions were: 10% mobile phase A (hold for 2 min) to 90% mobile phase B (hold for 10 min) over a 8-min gradient, with a 5-min equilibrium between runs. Mobile phase A: 82% methanol/ 18% acetonitrile/0.1% formic acid; mobile phase B: 99% water/0.1% formic acid. The average recoveries from PLE from the spiked biosolids (Milorganite® and Hyperion biosolids), were as follows: roxithromycin 13% (n=6), clarithromycin 40% (n=6), and azithromycin 24% (n=6). However, when comparing the results between the two biosolids materials the recoveries are different: Milorganite®: roxithromycin 19% (n=4), clarithromycin 54% (n=4), and azithromycin 28% (n=4); Hyperion biosolids: roxithromycin 1% (n=2), clarithromycin 13% (n=2), and azithromycin 16% (n=2). We believe that this difference in recoveries is the result of the variations that are seen in the physical and chemical composition of each biosolids, e.g., varying levels of lipids, de-watering processes, chemical stabilizers/additives. Also, the macrolide antibiotics are very large and positively charged molecules, while the biosolids particulates have large interstitial spaces and negative charges. The results from the unspiked Class A biosolids are reported in Table 2. All three macrolides were detected in the Milorganite® sample. They ranged from 0.4 µg/kg (dry wt) for roxithromycin to 14 µg/kg for azithromycin. If we were to correct for extraction efficiency then their values would be 2 µg/kg for roxithromycin and 50 µg/kg for azithromycin. The two

macrolides, azithromycin and clarithromycin, were detected in the Hyperion biosolids, 25 µg/kg and 20 µg/kg, respectively (corrected values 152 µg/kg and 160 µg/kg, respectively). The azithromycin results are comparable to those found by Göbel et al. [32] in European biosolids. Also detected, and quantified, in the Hyperion biosolids was methamphetamine at 4 µg/kg (dry wt). This value for methamphetamine is similar to that reported by Kaleta et al. [34] for amphetamine at 5 to 300 µg/kg (dry wt). Amphetamine is a more commonly used illicit substance than methamphetamine in the EU. We believe that this is the first time that a drug whose major origin is from illicit use has been detected and reported in US biosolids.

Regulating Biosolids in the United States

The EPA promulgated the 40 CFR Part 503, *Standards for the Use or Disposal of Sewage Sludge*, in 1993 [1]. The 1993 rule established requirements for the final use or disposal of sewage sludge when it is: 1) applied to land as a fertilizer or soil amendment; 2) placed in a surface disposal site, including sewage sludge-only landfills; or 3) incinerated. The Part 503 rule resulted in numerical standards for ten metals and operational standards for reducing microbial organisms in biosolids and to reduce vector attraction.

The 40 CFR Part 503 also allows disposal of sewage sludge in a municipal solid waste landfill that meets the requirements of 40 CFR Part 258. Part 503 prescribes numerical limits for three metals in sewage sludge placed in surface disposal sites, and sets forth requirements for the placement and the management of sewage sludge in a surface disposal site.

The Agency has also established limits for five metal pollutants in sewage sludge to be incinerated in a sewage sludge incinerator (SSI) and adopted standards under the Clean Air Act for two additional metal pollutants. The performance standards for SSIs include an operational standard for total hydrocarbons or carbon monoxide emissions that control numerous organic compounds found in SSI emissions.

A technical basis of the Part 503 standards is a comprehensive risk assessment that produces standards protective of human health and the environment, and is dependent upon the appropriate input of toxicity and environmental data of sewage sludge pollutants. In this assessment, exposure and hazard assessments for potential pollutants in sewage sludge ideally utilize empirical data obtained from field studies. Field studies and probabilistic modeling using 14 exposure pathways allow a conservative exposure and hazard assessment for pollutants in sewage sludge. Figure 2 depicts the conceptual site model for the agricultural application of biosolids, the 14 exposure pathways, and the highly exposed individuals (the farm family).

To develop the risk assessment, the Agency assumes that farm families apply biosolids to cropland where exposed fruits, vegetables, and root crops grow, and to pasture land where beef and dairy cattle graze. The farm family, who applies biosolids, is assumed to consume a high percentage of their own farm-raised products. The Agency further assumes that farmers live on a small strip of land (the buffer area) between the crop or pasture and the stream. The farmer raises free-range chickens in a yard that is also located in the buffer area. Beyond the buffer area is a fishable third-order stream. The farmer, his lactating wife, their infant, and older children may come in contact with pollutants via the 14 exposure routes, which include: 1) plants grown in biosolids-amended soils; 2) livestock grazing in treated areas; 3) consumption of crops grown in treated areas; 4) consumption of meat and milk from livestock that graze in the treated areas; 5) wildlife living, feeding and foraging in treated areas; 6) inhalation of ambient air; and 7)

consumption of drinking water from an index reservoir, or groundwater, that are potentially contaminated from pollutants released from biosolids.

As mentioned previously, EPA commissioned the NRC of the National Academy of Sciences (NAS) to independently review the technical basis of the chemical and microbial regulations applicable to sewage sludge applied to land. In July 2002, the NRC published a report entitled *"Biosolids Applied to Land: Advancing Standards and Practices"* in response to the EPA's request [4]. The NRC Committee concluded "There is no documented scientific evidence that the Part 503 rule has failed to protect human health. However, additional scientific work is needed to reduce persistent uncertainty about the potential for adverse human health effects from exposure to biosolids." The NRC identified a need to update the scientific basis of Part 503 and provided approximately 60 recommendations.

In April 2003, EPA announced and requested public comments on a preliminary strategy explaining how EPA planned to respond to the NRC report [4] on biosolids and the recommendations therein [35]. On 31 December 2003, the Agency announced its final response strategy, also known as the Final Action Plan [36]. EPA's final strategy in the *31 December 2003 Federal Register Notice* indicated that while emphasis was being placed on pathogens to address areas of uncertainty and public interest, selected new chemicals would also be addressed to help determine significant issues and identify information gaps that remain to be addressed [36]. Some PPCPs are among those chemicals that EPA intends to study.

The NRC Report specifically identified PPCPs as one category of diverse compounds that had not been studied in biosolids and that is especially likely to be present in domestic biosolids. The NRC report indicated that there is a need for a new hazard assessment of biosolids and to expand the suite of chemicals to be evaluated. EPA's Office of Research and Development is developing chemical analysis methods for certain PPCPs (e.g., macrolide and fluoroquinolone antibiotics and synthetic musks). These methods will be adapted for biosolids and converted to 40 CFR Part 136 methodologies. Subsequently, the Agency may apply these methods to a limited number of real-world samples for a pilot-scale survey of PPCPs in biosolids. For example, starting in late 2006 EPA will conduct a targeted national sewage sludge survey from 80 randomly selected WWTPs from across the US. The potential list of targeted chemicals includes several antibiotics, drugs, steroids, and hormones.

European Union Regulatory Aspects of Sewage sludge

Regulations in the U.S. and in the European Union (EU) share the same objective of controlling pathogens and pollutants in sewage sludge, although differences exist in specific requirements, not only between the EU and the US, but also among the EU member countries. Despite regulations to reduce the risk from sewage sludge, public opposition to sewage sludge land application is growing in the EU, just as it is in the US. Wastewater treatment facilities and sewage sludge producers face increasing difficulty in using and disposing of sewage sludge.

Government agencies in the EU have issued regulations on the land application of sewage sludge, seeking to limit the risks from pathogens and pollutants [37]. Regulations regarding the agricultural use of sewage sludge in the EU are described in a 1986 Directive containing 18 articles [38].

Currently, the EU consists of 15 member countries, mostly located in Western Europe. Expansion to 21 countries is likely to occur in the near future, while other countries, mostly from the Eastern region of the EU, are considering membership. The land area presently occupied by the EU is smaller than that of the U.S., but its population is larger. The average population density in the EU is four times greater than in the US. Therefore, sewage sludge management may be a more urgent issue in the EU, since Europe produces more sewage sludge and has less agricultural area available for recycling of the material [37].

Similar to the US, individual member countries are allowed to adopt standards more stringent than those established by the EU. In general, the standards from the Netherlands are the strictest [37]. Some individual countries have adopted lower heavy metal limits, or have included limits for pathogens or organic pollutants. For example, Belgium, Denmark, Finland, the Netherlands, and Sweden have lower heavy metal limits than the 1986 EU Directive, and Austria, Belgium, Denmark, France, Germany, and Sweden have organic compounds limits.

Considering the differences in standards among member and neighboring EU countries, waste flows have been observed in countries imposing more stringent standards (France and Belgium) from those countries with more lenient measures (the Netherlands, Germany, and Switzerland) [39]. For example, household waste is subject to conflicting interests that can highlight the clash of different national definitions of hazardous waste. In Germany, hospital waste is in the same category as household waste. Germany considers these non-hazardous and hospital and household waste can therefore circulate without restrictions. By contrast, France considers hospital waste hazardous, and prohibits its entry into the country [39].

The EU 1986 Directive does not specify limits for pathogen densities, but requires treatment of sewage sludge prior to land application in order to reduce pathogen densities unless the sewage sludge is injected or incorporated into the soil [38]. Requirements for sewage sludge treatment are the responsibility of individual member states. For instance, the treatment processes adopted by the UK are comparable to the U.S.'s "Processes to Significantly Reduce Pathogens" (PSRPs) for sewage sludge Class B production [40]. These include such processes as aerobic digestion, composting and lime stabilization. Restrictions on the application of sewage sludge on farmland exist, depending on the purpose of the land and/or the agricultural crop. Sewage sludge produced in an advanced treatment process have few restrictions regarding land application, whereas conventional treatments produce sewage sludge that has more limited applicability [37].

The EU is seeking to promote sewage sludge land application by reducing potential risks, by further research, and by increasing public confidence in the safety of the product [37,41]. Only a few studies have been performed on organic compound concentrations in sewage sludge, and a full evaluation is further hampered by the fact that, at present, no universally accepted and validated chemical analytical methods exist for analyzing most organic compounds [37,42], including PPCPs, in sewage sludge.

Decreasing the risk of pathogens in sewage sludge has so far been accomplished by reducing pathogens through Class A treatment technologies (e.g., high temperature or high pH), or by observing Class B harvesting and grazing restrictions. Decreasing the risk from pollutants has so far been accomplished by controlling pollutants at the source through pretreatment programs. Source control technologies, and reduction of use, have led to decreasing

concentrations of certain pollutants (e.g., phthalates, nonylphenol, polyaromatic hydrocarbons and dioxins) in sewage sludge over the past years [43,44]. Two classes of chemicals, 4-nonylphenols and phthalates, were reportedly reduced from sewage sludge during composting [45].

Conclusions

Some of the biggest analytical challenges to a "complete" analysis of sewage sludge include overcoming the large negative surface charges and interstitial spaces that provide multiple active sites for charged compounds, and the clean-up step for removing the bulk material (e.g., fats, proteins, surfactants) that are co-extracted with the pharmaceuticals. Certainly major chemical differences could be expected between the various sewage sludge products from both within the U.S., and between the U.S. and Europe. For example, regulations and treatment processes vary, prescribing practices differ in the U.S. and Europe, the populations contributing to each WWTP that produces sewage sludge can range from the hundreds to millions of individuals, the amount of interfering materials (e.g., surfactants, personal care products, lipids) present can differ from one cultural center to another, and the amounts of pharmaceuticals recovered from each type of material varies widely, as seen in Table 2. All of these facts point out that each sewage sludge material will have unique analytical challenges that will need to be addressed before final chemical signatures can be assigned. An even more important question that should be asked is whether these pharmaceutical residues are bioavailable, and if so, then what will be the environmental impact. Many of the challenges facing implementation of residuals management are the same, irrespective of the country of origin. Public consultation at early stages of residuals management will help countries implement environmentally acceptable programs. For example, a survey of households in the U.K. yielded information on when and how they disposed of unused pharmaceuticals [46]. Based on this information, the authors constructed a conceptual model to assess the pathways of human pharmaceuticals into the environment. The model demonstrated that disposal of unused pharmaceuticals, either by household waste or via the sink or toilet, may be a prominent route to the environment that requires greater consideration.

The introduction of new chemicals, and increased use of others, may result in their presence in sewage sludge. Brominated diphenyl ethers (flame retardants), nitro musks (synthetic perfumes), linear alkylbenzene sulfonates (detergents), pharmaceutical compounds (antibiotics and drugs), odorants (for sewage sludge odor control), and polyelectrolytes (for sewage sludge dewatering) are important targets of research as emerging pollutants of potential significance in sewage sludge both in Europe and the US [4,37,44].

Although concerns have been raised, no scientific evidence exists that the current practice of land application of sewage sludge is harmful either to human health or to the environment. The current standards for sewage sludge in the U.S., and in some EU member countries, are science-based risk assessments. With respect to pollutants in sewage sludge, particularly emerging contaminants such as PPCPs, more accurate data on use, advances in chemical analytical methodology, survival efficiencies in wastewater treatment facilities, environmental fate and transport, and the potential for effects in humans and the environment will be required to conduct reliable exposure and hazard assessments. Sufficient data to conduct an exposure and hazard assessment include unbiased national estimates of concentrations, environmental fate and transport, plausible effects end-points for humans and ecological receptors, and other relevant information for pollutants in sewage sludge.

Future U.S., and EU, legislation on the land application of sewage sludge may become more complex with new scientific and technological advancements. Future standards should continue to be based on sound science.

Acknowledgements
One of us (TLJ-L) would like to thank her technical Senior Environmental Employee, Mr. Tom Moy, for all of his hard work and assistance in the day-to-day laboratory operations.

Notice: The US Environmental Protection Agency (EPA), through its Office of Research and Development (ORD), funded and performed the research described. It has been subjected to the Agency's administrative review and approved for publication as an EPA document. Mention of trade names or commercial products does not constitute endorsement or recommendation by EPA for use.

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Table 1
MS/MS Product ions for identifying and quantifying

Analyte [CASRN]	Precursor ion	Product ion(s) with % relative abundance > 5%; [CE %]
Azithromycin [83905-01-5]	749.4 $(M+H)^+$	591.3 $(M+H-C_8H_{16}O_2N)^+$ [30] 573.3 $(M+H-C_8H_{16}O_2N-H_2O)^+$ 434.3
Clarithromycin [81103-11-9]	748.2 $(M+H)^+$	590.1 $(M+H-C_8H_{16}O_2N)^+$ [30] 558.2 $(M+H-C_8H_{16}O_2N-CH_3OH)^+$
Roxithromycin [80214-83-1]	859.4 $(M+Na-H)^+$	755.4 $(M+Na+H-C_4H_9O_3)^+$ [30] 597.3

Table 2

Pharmaceutical or Personal Care Product	Reference	Media	Extraction methodology	Detection methodology	Amount Detected Dry weight µg/kg
Synthetic Musks HHCB (Galaxolide) AHTN (Tonalide)	Stevens et al. [20]	sewage sludge digested	Soxhlet with GPC cleanup	Gas chromatography with mass spectrometry (GC-MS) - selected ion monitoring (SIM)	26,000 (HHCB) 4,000 (AHTN)
	Osemwengie [21]	biosolids - Class A Milorganite	Pressurized liquid extraction (PLE) with GPC cleanup	GC-MS-SIM	5,000 (HHCB) 2,000 (AHTN)
		Los Angeles Hyperion			18,000 (HHCB) 4,000 (AHTN)
		Las Vegas WWTP (Class B)			10,000 (HHCB) 3,000 (AHTN)
	Kinney et al. [33]	biosolids class A and sludge	PLE with post-extraction cleanup	GC-MS full scan	13 - 177000 (HHCB) 78 - 427000 (AHTN)
Fluoroquinolone(s) Ciprofloxacin (CP) Norfloxacin (NF)	Golet et al. [30]	sewage sludge raw	PLE with post-extraction cleanup using solid phase extraction (SPE)	Liquid chromatography-fluorescence detection (LC-FLD)	1000 - 2000 CP 1500 - 2000 NF
		sewage sludge digested			2300 - 2400 CP 2100 - 2400 NF
Methamphetamine	Jones-Lepp and Stevens (this work)	Biosolids - Class A Milorganite	PLE with post-extraction hexane cleanup	LC-electrospray-mass spectrometry/mass spectrometry (LC-ESI-MS/MS), positive ionization	0 (METH)
		Los Angeles Hyperion			4 (METH)
Amphetamine	Kaleta et al. [34]	Sewage sludge raw and treated	Re-suspension of solids into liquid matrix then SPE.	LC-ESI-MS/MS, positive ionization	5 - 300 (AMPH)

Macrolide antibiotics Azithromycin (AZI) Clarithromycin (CLA) Roxithromycin (ROX) Tylosin (TY) Erythromycin (ERY)	Göbel et al. [32]	sewage sludge (activated and digested)	PLE and USE with post-extraction cleanup with SPE	LC-electrospray-mass spectrometry/mass spectrometry (LC-ESI-MS/MS), positive ionization	1.3 - 158 (AZI) 0.3 - 63 (CLA) nd - 131 (ROX)
	Jacobsen et al. [29]	swine manure	PLE with post-extraction cleanup using liquid-liquid extraction followed with SPE	LC-ESI-MS/MS, positive ionization	nd (TY)
	Jones-Lepp and Stevens (this work)	Biosolids - Class A Milorganite Los Angeles Hyperion	PLE with post-extraction cleanup with hexane	LC-ESI-MS/MS, positive ionization	14 (AZI) 9 (CLA) 0.4 (ROX) 25 (AZI) 20 (CLA) nd (ROX)
	Kinney et al. [33]	biosolids class A & B, and sludge	PLE with post-extraction cleanup	LC-ESI-MS, positive ionization	nd - 41 (ERY)
Tetracyclines Tetracycline (TT) Oxytetracycline (OT) Chlortetracycline (CT) Doxycycline (DXY) Epi-tetracycline (ETT) Epi-oxytetracycline (EOT) Epi-chlortetracycline (ECT)	Jacobsen et al. [29]	swine manure	PLE with post-extraction cleanup using liquid-liquid extraction followed with SPE	LC-ESI-MS/MS positive ionization	nd - 1600 TT nd - 1500 OT nd - 16,000 CT 550 - 3100 DXY nd - 990 ETT nd - 450 EOT nd - 14,100 ECT
Trimethoprim	Göbel et al. [32]	sewage sludge (activated and digested)	PLE and USE with post-extraction cleanup with SPE	LC-ESI-MS/MS, positive ionization	nd - 133
	Kinney et al. [33]	biosolids class A & B, and sludge	PLE with post-extraction cleanup	LC-ESI-MS, positive ionization	nd - 22

Sulfonamides Sulfadiazine (SD) Sulfamethazine (SM) Sulfadoxine (SDX) Sulfamethoxazole (SMZ) Sulfapyridine (SPY)	Jacobsen et al. [29]	swine manure	PLE with post-extraction cleanup using liquid-liquid extraction followed with SPE	LC-ESI-MS/MS positive ionization	nd - 2100 SD nd SM nd - 220 SDX
	Göbel et al. [32]	sewage sludge (activated and digested)	PLE and USE with post-extraction cleanup with SPE	LC-ESI-MS/MS, positive ionization	nd - 113 SMZ nd - 197 SPY
	Kinney et al. [33]	biosolids class A and sludge	PLE with post-extraction cleanup	LC-ESI-MS, positive ionization	nd - 160 SMZ
Triclocarban (topical antiseptic)	Heidler et al. [31]	sewage sludge digested	PLE	LC-ESI-MS/MS (SIM) negative ionization	51,000
Other pharmaceuticals					
acetaminophen albuterol carbamazepine dehydronifedipine diltiazem fluoxetine gemfibrozil thiabendazole warfarin cimetidine codeine diphenhydramine miconazole	Kinney et al. [33]	biosolids class A and sludge	PLE with post-extraction cleanup	LC-ESI-MS, positive ionization	nd - 1400 nd - 850 8 - 390 nd - 26 nd - 59 nd - 1500 nd - 420 nd - 5000 nd - 92 nd - 71 nd - 22 15 - 7000 nd - 460

Figures

Figure 1

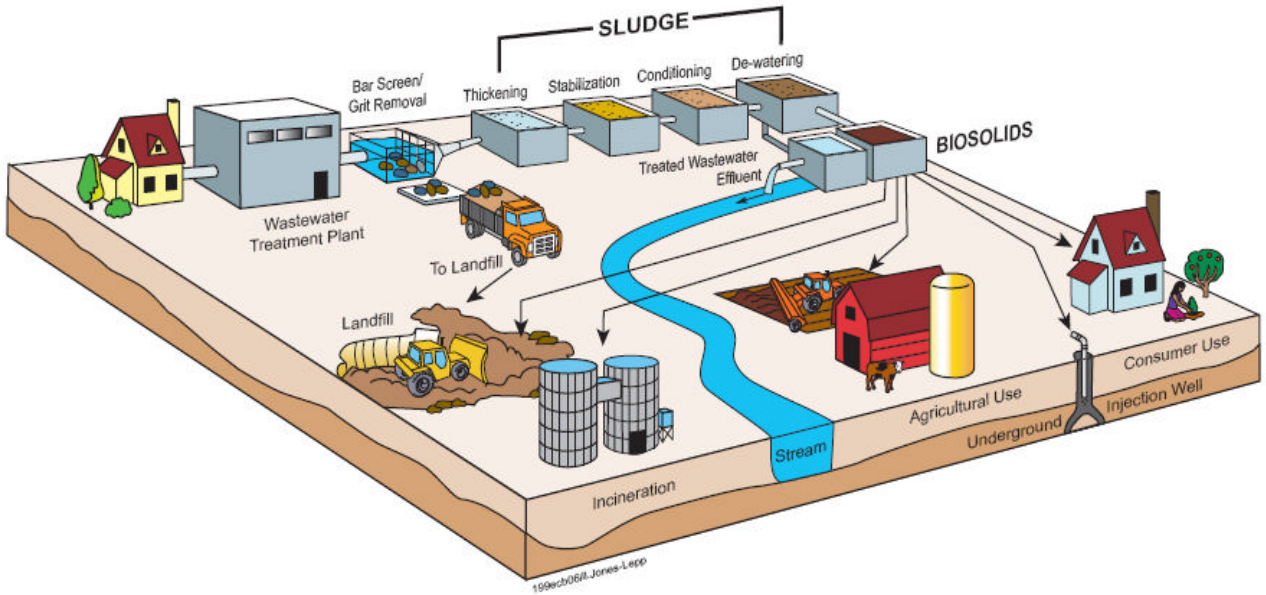
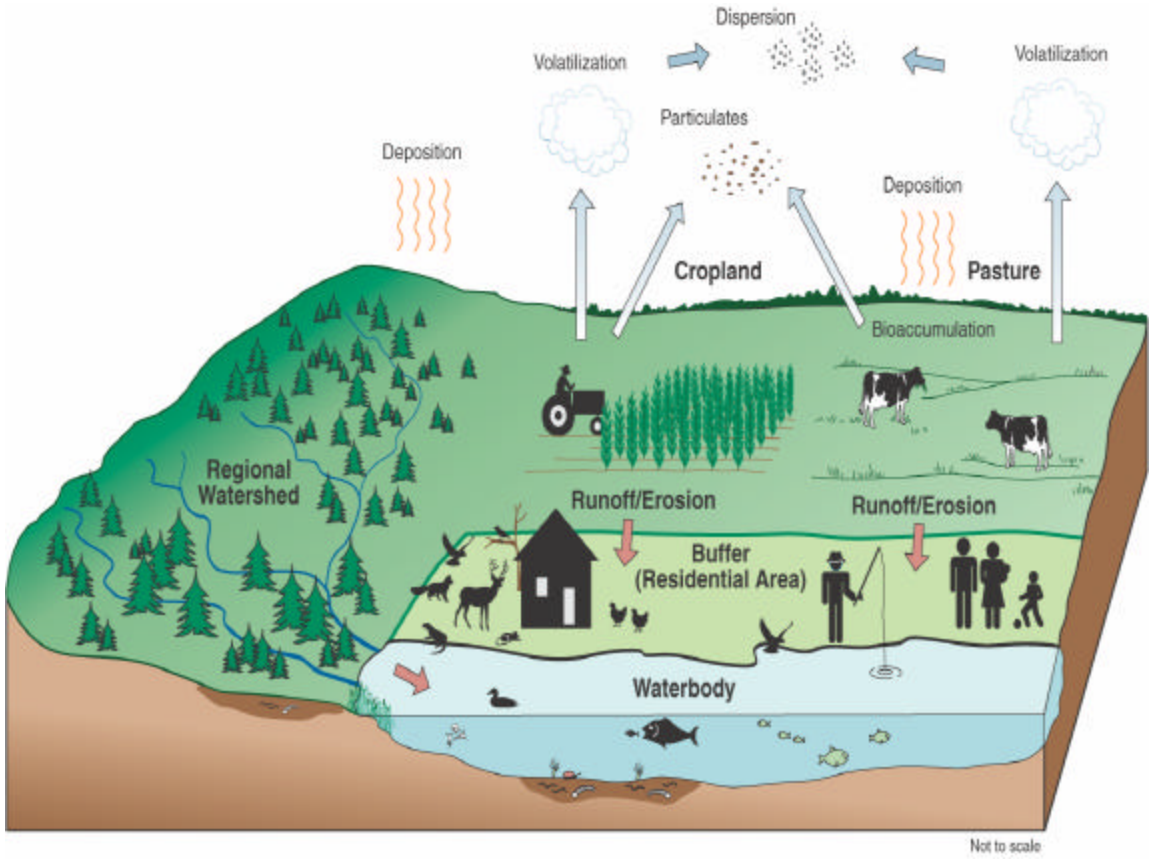

SLUDGE
Bar Screen/
Grit Removal
Thickening
Stabilization
Conditioning
De-watering
BIOSOLIDS
Treated Wastewater
Effluent
Wastewater
Treatment Plant
To Landfill
Landfill
Consumer Use
Agricultural Use
Injection Well
Underground
Stream
Incineration
199ech06it.Jones-Lepp

Figure 2


Dispersion
Volatilization
Volatilization
Particulates
Deposition
Deposition
Cropland
Pasture
Bioaccumulation
Regional Watershed
Runoff/Erosion
Runoff/Erosion
Buffer (Residential Area)
Waterbody
Not to scale